REMARKS

Applicant respectfully requests reconsideration and allowance of the subject application.

Claims 1-2, 4-5, 8-9, 11, 16, 21-23, 51-53, 71-72, and 74-80 were previously pending.

Please cancel claims 51-53, and 78-80.

Please amend claims 1, 4-5, 8-9, 11, 16, 71-72, and 75 as shown below.

Claims 1-2, 4-5, 8-9, 11, 16, 21-23, 71-72, and 74-77 are currently pending.

Claim Rejections under 35 U.S.C. § 102

The Office rejected claims 71, 72, and 74 under 35 USC § 102(a) as being anticipated by Gordon (U.S. Patent No. 6,607,911); and under 35 USC § 102(b) as being anticipated by Chenchik (U.S. Patent No. 5,759,822).

The Gordon Reference

Gordon produces a pure (cassette specific) collection of double-stranded fragments for each single cassette that is eventually used for digestion and subsequent ligation 5' to 3' into a construct containing a vector.

The Gordon methodology generates *double-stranded* fragments that are then *ligated* into a *vector* to produce a single control molecule carrying multiple mutations "in series". Applicant's methodology, in contrast, produces small fragments that are *single-stranded* and amplify *exponentially* by either an internal or external primer pair based on the design of the artificial tags introduced at the

5' and 3' end of each fragment. Thus, Applicant's control carries multiple control molecules "in parallel."

The Chenchik Reference

The Chenchik reference discloses a method for producing oligonucleotide fragments to modulate amplification through suppression. The method is not, and does not work as, a reference solution. The Chenchik method uses complementing "adapter" regions on a single-stranded oligonucleotide that form loop structures in the absence of complementary fragments that do not amplify by PCR. The Chenchik reference requires interaction with the sample of interest and improves upon current, commonly used, known testing practices. The Chenchik method is used to enhance methods to test for the presence of different mutations and genetic conditions, but does not produce a product that emulates the mutations themselves.

Claim 71

Claim 71, as amended, defines a method for tuning concentrations of different reference nucleic acids within a clinical reference solution, including:

- designing multiple reference nucleic acids, wherein each reference nucleic acid comprises an arrangement of bases emulating a clinically relevant site on genes responsible for human genetic conditions exclusive of clinically irrelevant human nucleic acid adjacent to the clinically relevant site in vivo;

- for each reference nucleic acid in a first subset of the multiple reference nucleic acids, constructing an oligonucleotide comprising an arrangement of bases to emulate the clinically relevant site as isolated from clinically irrelevant nucleic acid that occurs adjacent to the corresponding clinically relevant site in vivo, including designing two ends of the arrangement to form a first pair of primer targets allowing PCR amplification of the first subset via a primer set specific to the first pair of primer targets;

- for each reference nucleic acid in a second subset of the multiple reference nucleic acids, constructing an oligonucleotide comprising an arrangement of bases to emulate the clinically relevant site as isolated from clinically irrelevant nucleic acid that occurs adjacent to the corresponding clinically relevant site in vivo, including designing two ends of the arrangement to form a second pair of primer targets allowing PCR amplification of the second subset via a second primer set specific to the second pair of primer targets; and

- wherein each oligonucleotide in the first and second subsets is constructed base-by-base, from end to end, as a single strand via a non-ligase, non-template, non-cloning synthesis.

Gordon and Chenchik do not show or disclose each element of Applicant's claim 1. Neither Gordon nor Chenchik show or disclose designing and constructing mixtures of oligonucleotides that can be individually and differentially amplified within the mixture, in which each oligonucleotide emulates a site on a gene while excluding extraneous (clinically irrelevant) nucleic acid adjacent in vivo to the clinically relevant site.

Since neither Gordon nor Chenchik show or describe each element of claim 71, as required for a 35 USC § 102 rejection, Applicant respectfully requests that the 35 USC § 102 rejections be removed from claim 71, and suggests that claim 71 is allowable.

Claims 72 and 74

For at least the reasons described above with respect to claim 71, in addition to their own content, claims 72 and 74 are also allowable. Dependent claims contain the language of their respective base claims. Claims 72 and 74 depend from claim 71. Thus, claims 72 and 74 are also allowable.

Claim Rejections under 35 U.S.C. § 103(a)

Claims 1, 2, 4-5, 8-9, 11, 16, 21-23, and 75-80 were rejected under 35 U.S.C. § 103(a) as being unpatentable over Gordon in view of Strizhov (U.S. Patent No. 6,110,668).

Of these rejected claims, claims 78-80 have been canceled.

The Strizhov Reference

Strizhoz describes a method of gene synthesis that permits codons of a natural gene to be changed to allow preferential transcription and translation of the synthetic gene in transgenic organisms. The method uses an enzymatic and chemical synthesis to create phosphorylated oligonucleotides of the gene, which are then assembled on a single-stranded partially homologous template DNA derived from the natural of wild-type gene. After annealing, the nicks between adjacent nucleotides are closed by a thermostable DNA ligase using repeated cycles of melting, annealing, and ligation. This template-directed-ligation ("***TDL***") results in a new single-stranded synthetic DNA product which is subsequently amplified and isolated from the wild type template strand by PCR with short flanking primers that are complementary only to the new synthetic strand. The PCR end-primers contain suitable restriction cleavage sites for cloning of the synthetic double-stranded DNA fragments (column 2, lines 45-65).

Claim 1

Claim 1, as amended, defines a method of creating a clinical reference solution that emulates clinically relevant sites on genes responsible for human genetic conditions, wherein the clinical reference solution is substantially free of clinically irrelevant nucleic acid, including:

- for each clinically relevant site, designing an oligonucleotide comprising an arrangement of bases to emulate the clinically

relevant site as isolated from clinically irrelevant nucleic acid that occurs adjacent to the corresponding clinically relevant site in vivo, including designing two ends of the arrangement to form primer targets for differentially amplifying the emulated clinically relevant site;

- for each arrangement, performing a non-ligase, non-template, non-cloning synthesis that includes constructing base-by-base, from end to end, a single strand of bases comprising the arrangement of bases that emulates the clinically relevant site and forms the primer targets associated with the clinically relevant site; and

- mixing each single strand into a single solution to form a collection of oligonucleotides each representing a clinically relevant site of a gene.

The Gordon reference and the Strizhov reference, singly or in combination do not teach or suggest each element of claim 1.

A person skilled in the art would not be motivated or otherwise informed by the references themselves to combine Gordon and Strizhov to arrive at Applicant's claim 1. That is, a person skilled in the art would not apply Strizhov's template-directed-ligation "*TDL*" for assembling a gene (upon a single-stranded partially homologous template DNA derived from the natural or wild-type gene) to Gordon's methodology for generating double-stranded fragments that are then

ligated into a vector to produce a single control molecule carrying multiple mutations in series, in order to arrive at the elements of Applicant's claim 1. In claim 1, a clinically relevant site on a gene, such as a mutation site, is emulated by an oligonucleotide. The entire gene is not emulated or constructed. Oligonucleotides are short base sequences. In the method of Applicant's claim 1, no template is needed to assemble a sequence longer than an oligonucleotide to represent the base sequence of the clinically relevant site (on the larger gene). Those parts of a gene that are beyond the clinically relevant site of interest are intentionally excluded from the oligonucleotide provided by claim 1, while Strizhov aims to produce an entire gene.

Strizhov utilizes oligonucleotides in a first stage of the Strizhov *TDL* procedure (see stage 1 in Strizhov's Fig. 1) yet Strizhov's utilization of oligonucleotides teaches against Applicant's claim 1. Strizhov applies a chemical phosphorylation during the Strizhov automated oligonucleotide synthesis in the Strizhov first stage so that only the full-length oligonucleotides have a 5'-phosphate group. By this arrangement only phosphorylated (i.e., full-length) oligonucleotides can be ligated with other adjacent oligonucleotides during the subsequent *TDL* stages, thereby ***conveniently eliminating oligonucleotides that are too short*** (column 4, lines 5-11; emphasis added). Strizhov uses oligonucleotides that have lengths between 80-130 bases, each representing e.g., one-eighth of the gene to be assembled.

At no point in the descriptions do Strizhov and/or Gordon teach, suggest, or hint at an oligonucleotide that mimics or emulates only a clinically relevant site (e.g., mutation) on a gene, nor do these references teach or suggest that such an

oligonucleotide should be amplified individually in order to make the oligonucleotide itself a control molecule in a reference solution. Accordingly, Strizhov (and Gordon) do not teach or suggest an embodiment that, for example, ends at the first stage (shown in Fig. 1) of Strizhov's *TDL* procedure in order to provide an oligonucleotide that represents only a specific site on a gene. Nor do Strizhov and/or Gordon teach or suggest primer sequences on each end of such a site-specific oligonucleotide. Construction of such a clinical reference oligonucleotide with primer sequences seamlessly added on the 3' and 5' ends is recited only in Applicant's of claim 1.

The practical upshot is that Applicant's claim 1 results in more direct and easy manufacture of a clinical reference solution in which the oligonucleotides so purely represent the clinically relevant sites at controllable potencies that the clinical reference solution can test for 30, 40, 50 or more mutations, whereas conventional techniques can test for only approximately 5 mutations. Moreover, the concentration of each species of oligonucleotide in Applicant's clinical reference solution can be individually and precisely controlled over a continuous spectrum of concentrations. That is, the concentration of each type of oligonucleotide can be differentially and independently controlled. With conventional techniques, control of concentration is clouded by presence of extraneous nucleic acid materials that these techniques inevitably introduce or maintain. Conventional techniques trade-off purity of the reference against ability to control concentrations.

Since neither Gordon nor Strizhov, alone or in combination, teach or suggest each element of claim 1 as amended, the combination fails. Applicant

respectfully requests that the 35 USC § 103 rejections be removed from claim 1, and suggests that claim 1 is allowable.

Claims 2, 4-5, 8-9, 11, 16, 21-23, and 75-77

For at least the reasons described above with respect to claim 1, in addition to their own content, claims 2, 4-5, 8-9, 11, 16, 21-23, and 75-77 are also allowable. Dependent claims contain the language of their respective base claims. Claims 2, 4-5, 8-9, 11, 16, 21-23, and 75-77 depend from claim 1. Thus, claims 2, 4-5, 8-9, 11, 16, 21-23, and 75-77 are also allowable.

Conclusion

The Applicant submits that all of the remaining claims are in condition for allowance and respectfully requests such allowance. If unresolved issues remain, Applicant respectfully requests that the undersigned attorney be contacted for scheduling an interview.

Respectfully Submitted,

Dated: March 27, 2008 By: /Mark Farrell/

Mark Farrell
Lee & Hayes, PLLC
Reg. No. 45,988
Attorney for Applicant
Telephone: 509-324-9256 x 243
Facsimile: (509) 323-8979